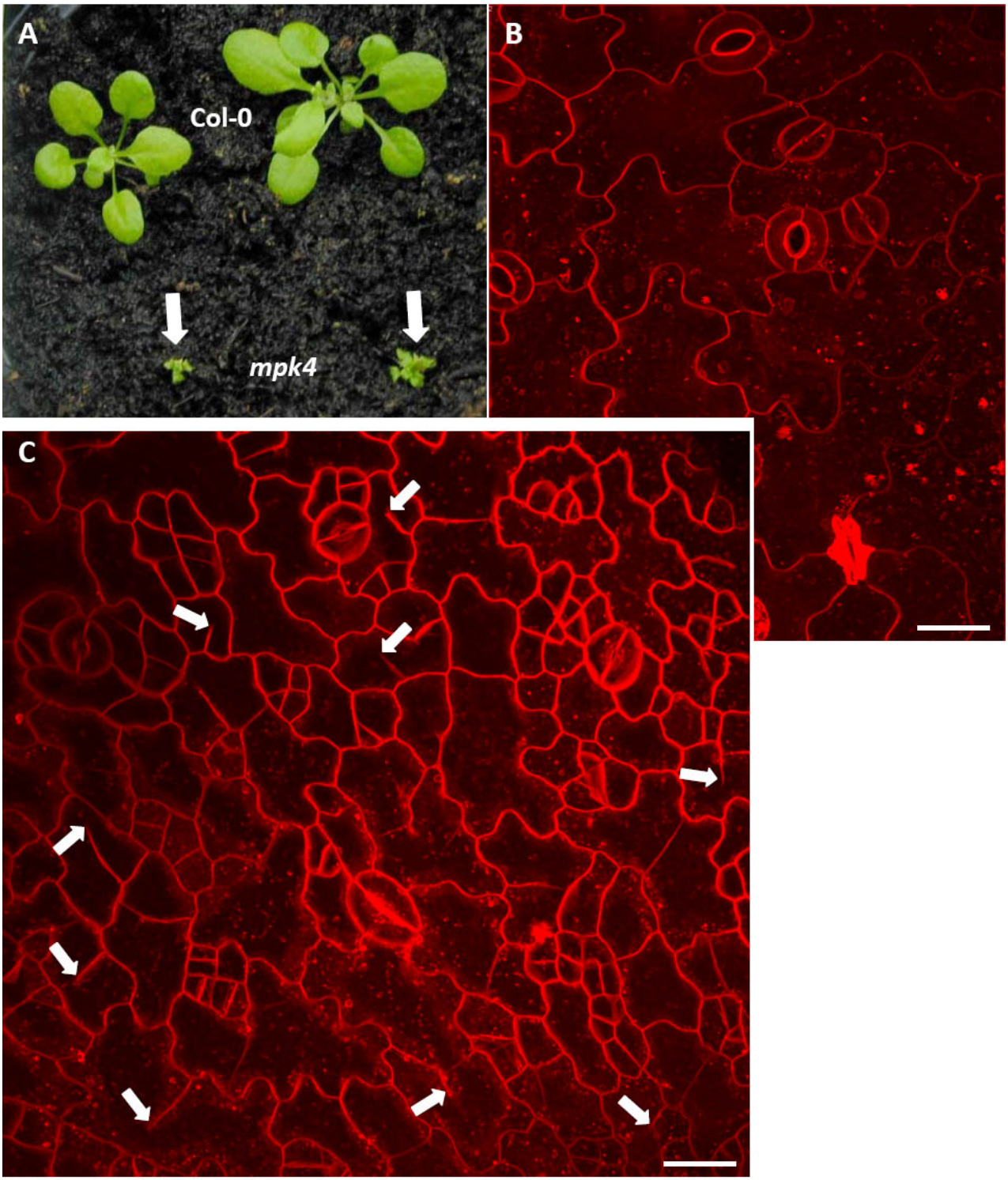
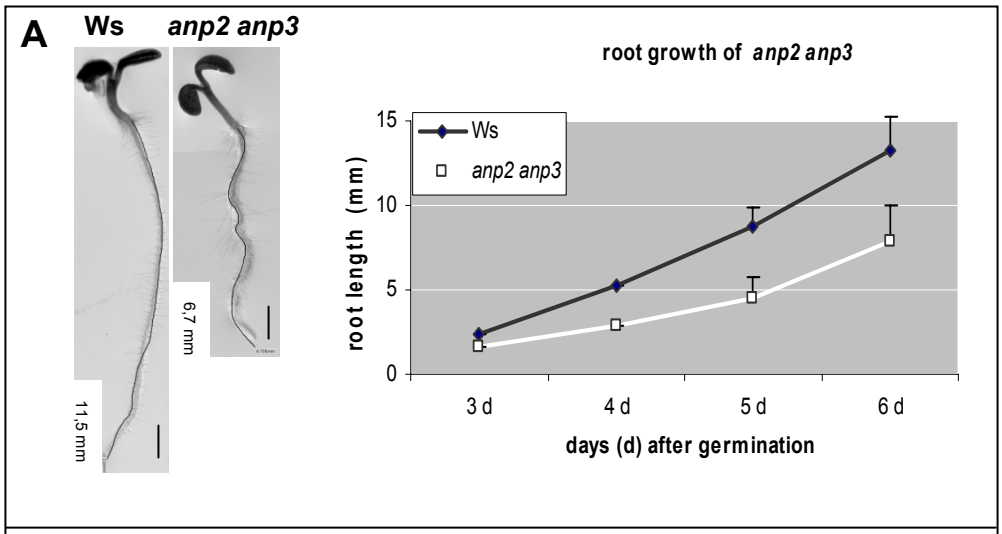


Supplemental Figure 1. Phenotypic comparison of the rosette leaves of four-week-old *mpk4* and Col-0 plants.
A *mpk4* vs Col-0 plants grown in soil. Note the extreme dwarfism of the *mpk4* plants (white arrows) compared to their Col-0 counterparts.
B and **C** Delineation of cell periphery of Col-0 (B) and *mpk4* (C) leaf cells with the membrane marker FM4-64. In the case of the *mpk4* leaf, pavement cell morphogenesis is disturbed while in many cells, incomplete cell walls are observed (white arrows), reminiscent of the cytokinetic phenotype reported for *anp2 anp3* (Krysan et al., 2002). Bars represent 20 µm in B and C.



C

	Ws	*anp2 anp3*	Col-0	*mpk4*
3 d	2,380 ± 0,299	1,684 ± 0,308		
4 d	5,225 ± 0,551	2,879 ± 0,896	5,849 ± 1,097	2,686 ± 0,764
5 d	8,765 ± 1,059	4,555 ± 1,20	11,2045 ± 2,083	4,691 ± 1,323
6 d	1,3311 ± 1,904	7,834 ± 2,193	17,967 ± 2,522	6,987 ± 2,233

n = 30

± = StD

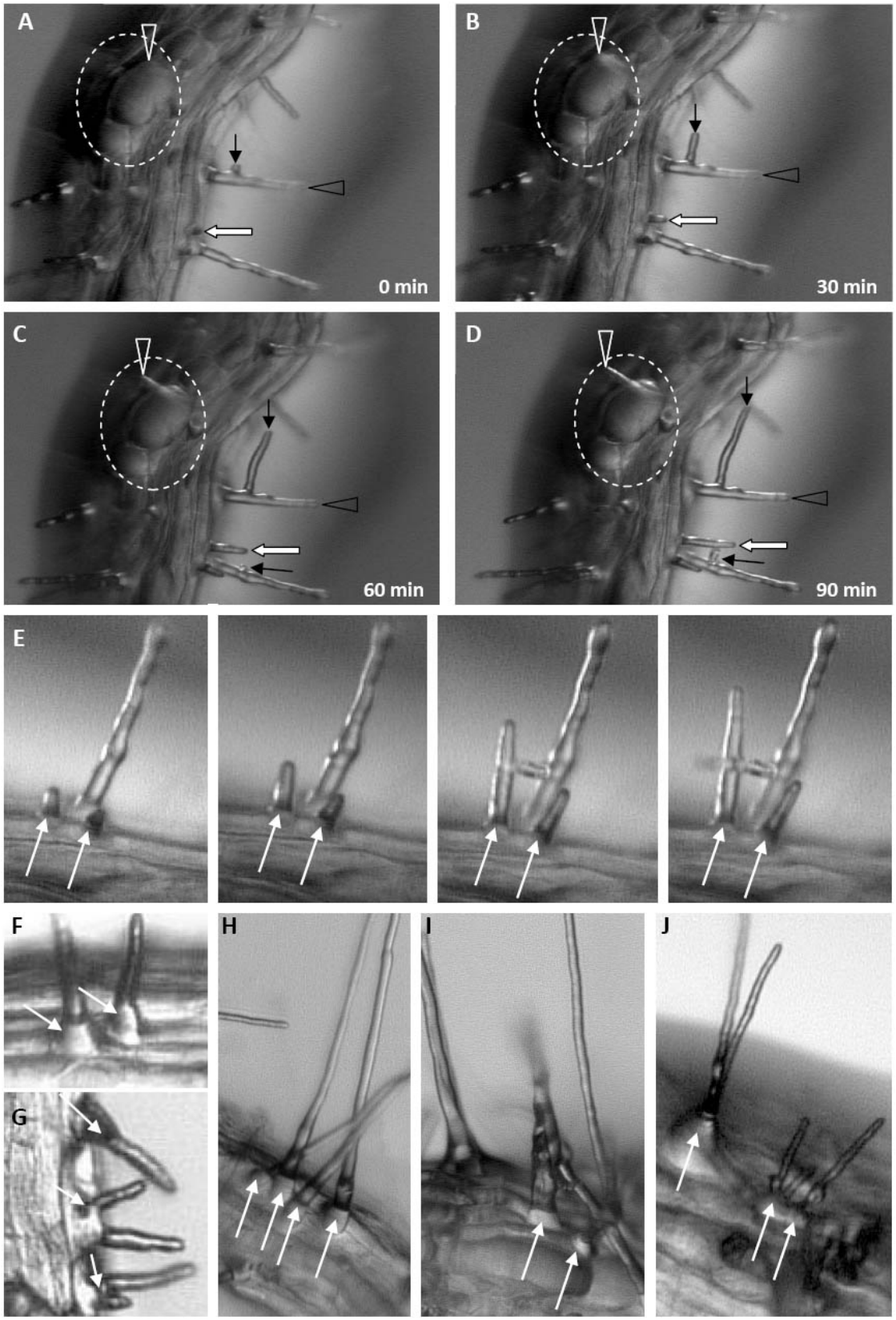
Supplemental Figure 2. Root growth of *anp2 anp3* and *mpk4* seedlings.

Lines along roots in representative images presented in A and B delineate root lengths which are shown in bottom corners in mm. Bars: 1mm.

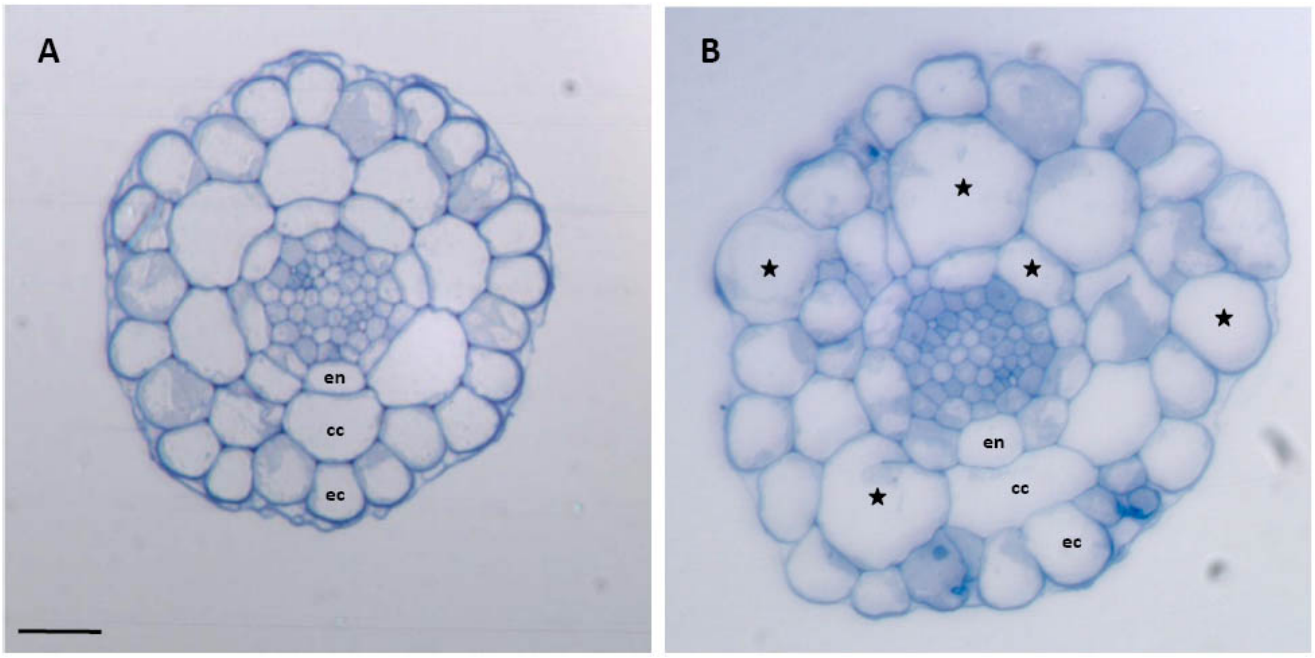
A Graphic depiction of root growth of a*np2xanp3* as compared to Ws. Mutant seedlings showing reduced growth. After six days of growth, the roots of the mutant plant they achieved only 2/3 of the length of roots from control plants.

B Graphic depiction of root growth of *mpk4* as compared to Col-0. Mutant seedlings showing retarded growth. The roots of the mutant plants reached only 1/3 of the length of control plants at 6th day.

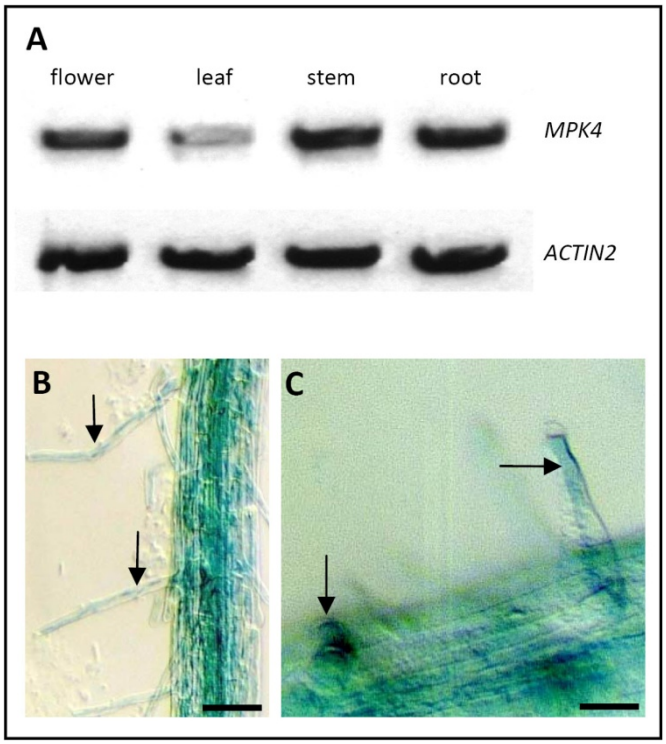
C Table showing the average root length in the mutants and corresponding wild types at different time points.



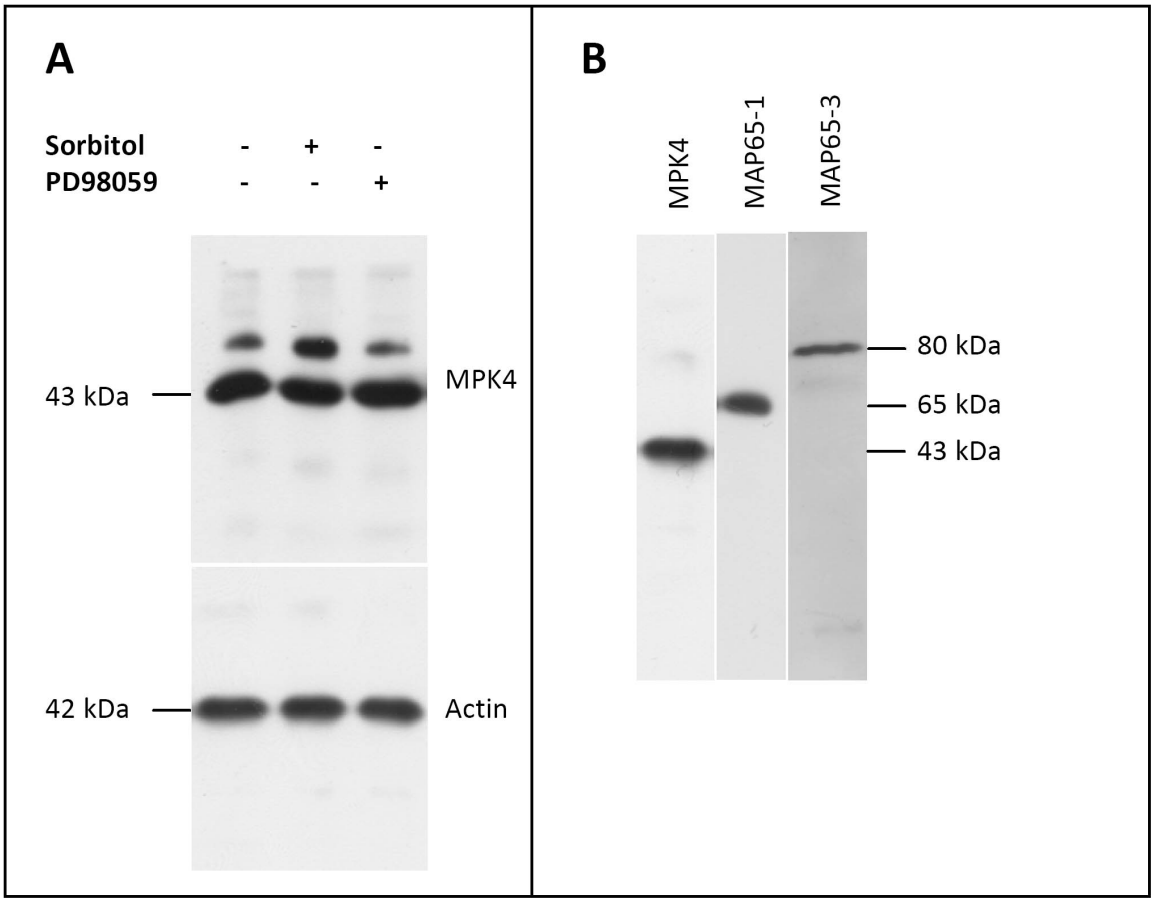
Supplemental Figure 3. Ectopic root hairs in *anp2 anp3* (A-G) and *mpk4* (H-J) mutants.
A-D Time-lapse imaging documenting root hair growth in 30 min intervals. Circle depict extremely expanded epidermal cell with outgrowing root hair. Black arrows point to outgrowing second root hair tips, while first root hair tip stops its growth (black arrowhead). White arrow points to ectopic root hair. **E** Detail of the video sequence shown in A-D. Ectopic root hairs are pointed by arrows. **F-J** More examples of ectopic root hairs (arrows) in both mutants.



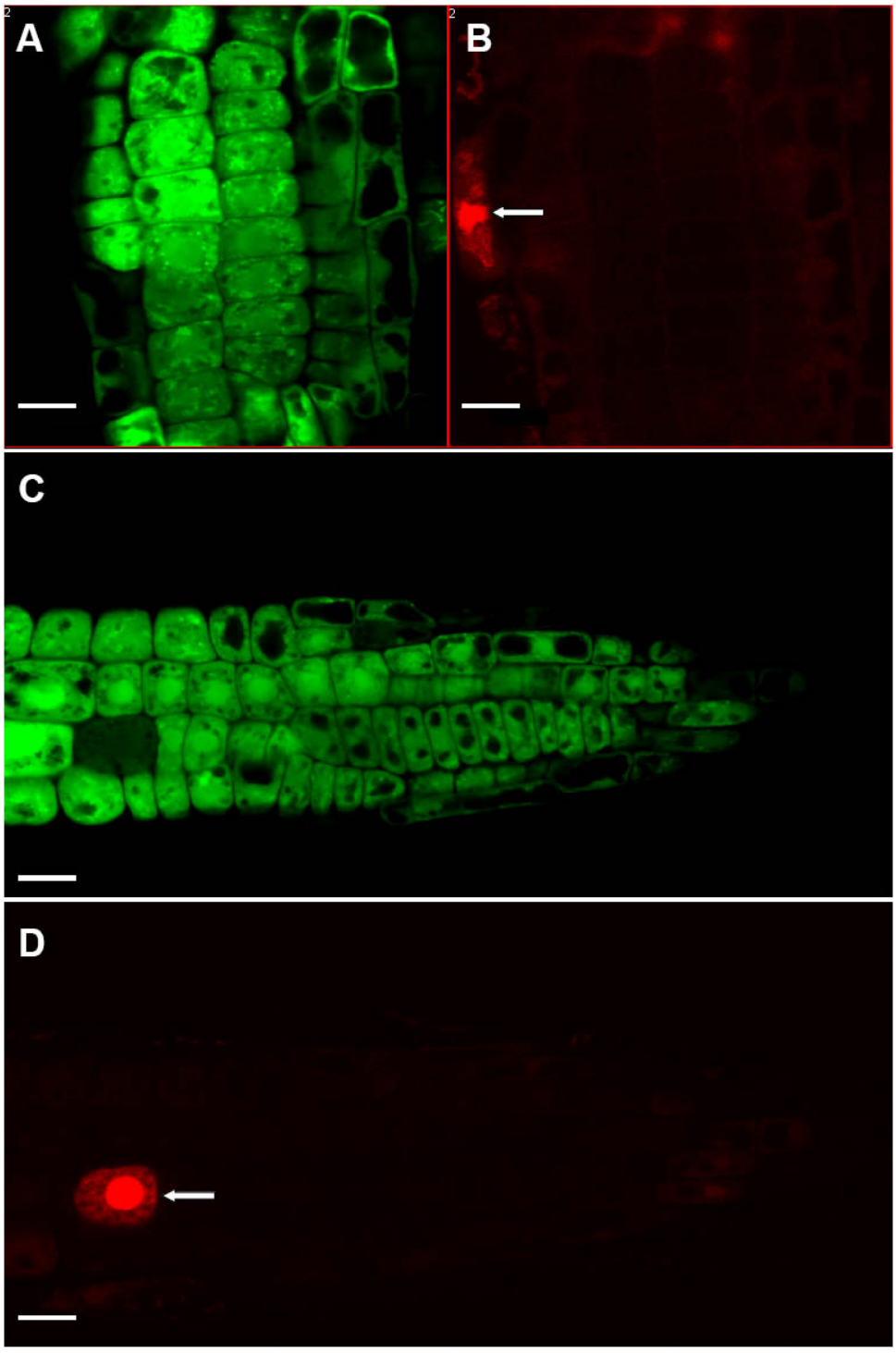
Supplemental Figure 4. Radial swelling of root cells in the *mpk4* mutant.
Comparative cross-sectional microscopic visualization of the root of Col-0 (A) and the *mpk4* (B) mutant. ec: epidermal cell, cc: cortical cell, en: endodermal cell. Asterisks denote cells showing prominent radial expansion in the *mpk4* mutant. Bar: 50 µm.



Supplemental Figure 5. Analysis of tissue- and cell-specific expression of *MPK4*.
A RT-PCR assessment (28 amplification cycles) of *MPK4* expression in flowers, leaves, stems and roots of Col-0 plants. *Actin 2* (28 amplification cycles) was used as a control. Three biological and three technical replicates were performed.
B and **C** Visualization (GUS histochemistry) of *MPK4* promoter activity in roots of *ProMPK4:GUS* stably transformed *A. thaliana* (Col-0) lines. *MPK4* promoter activity was found in root epidermal cells and roots within the root hair formation zone (B) while more detailed analysis showed the promoter activity in root hair bulges and root hairs (arrows in C). Bars: 100 µm in B and 20 µm in C.

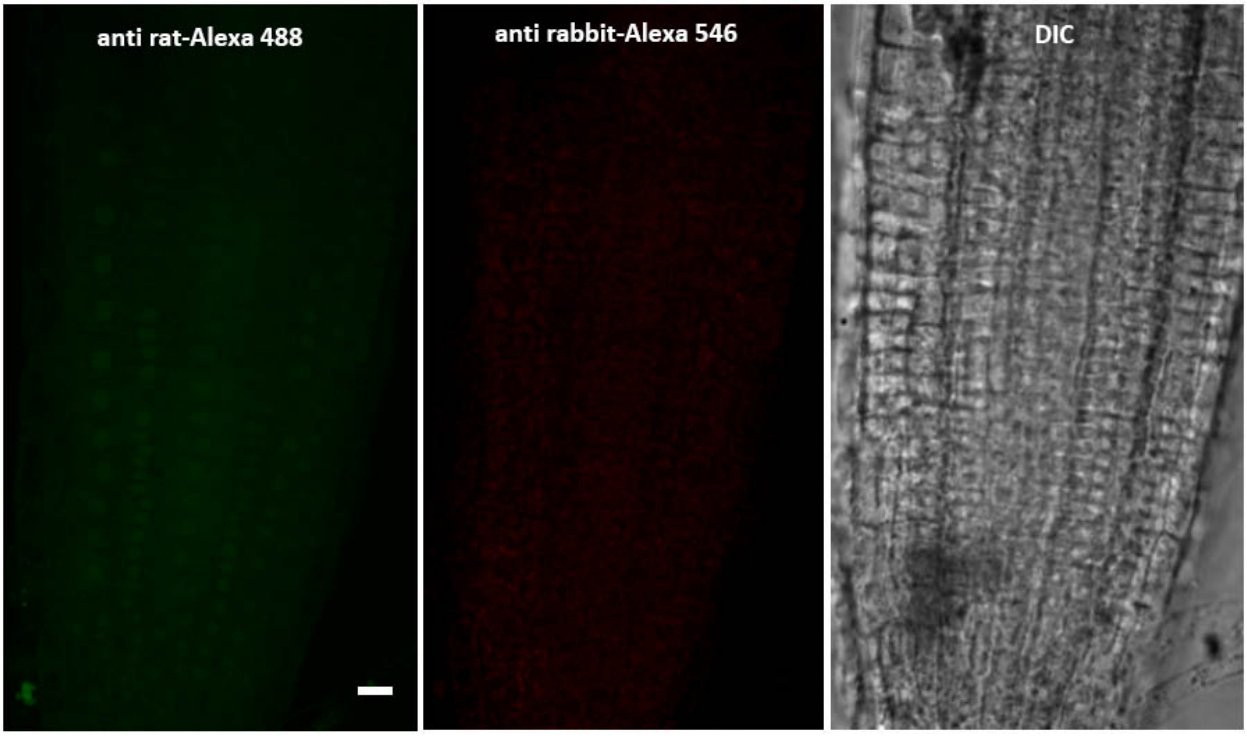


Supplemental Figure 6. Effect of PD98059 on MPK4 activation and specificity of MPK4, MAP65-1 and MAP65-3 antibodies.
A Monitoring of the activation of MPK4 through hyperosmotic sorbitol treatment by means of the Phos-Tag mobility retardation assay, and inhibition of the hyperosmotically-induced MPK4 activation by PD98059. **B** Immunoblots showing the specificity of the anti-MPK4, anti-MAP65-1 and the anti-MAP65-3 antibodies in *Arabidopsis* Col-0 extracts.



Supplemental Figure 7. Cell viability assessment in Arabidopsis roots using dual propidium iodide (PI)/ fluorescein diacetate (FDA) staining .

Control (A, B) and sorbitol-treated (C, D) root cells of Col-0. Dead cells are indicated by red nuclear fluorescence due to PI diffusion (arrows in B and D), whereas living cells are marked by green cytoplasmic fluorescence of FDA. Note that there is no obvious difference in the vitality of cells. Bar: 10 µm.



Supplemental Figure 8. Negative controls for immunofluorescent MAP65-1 and tubulin staining.
Root epidermal cells immunolabelled with only secondary antibodies were used as the negative control. Note that there is only very weak unspecific labelling of nuclei with the green fluorescent antibody. Bar: 10 µm.

Supplemental References

Krysan P.J., Jester P.J., Gottwald J.R., and Sussman M.R. (2002). An *Arabidopsis* mitogen-activated protein kinase kinase kinase gene family encodes essential positive regulators of cytokinesis. Plant Cell **14:** 1109-1120. PMID: 12034900